Saskatchewan
Agriculture

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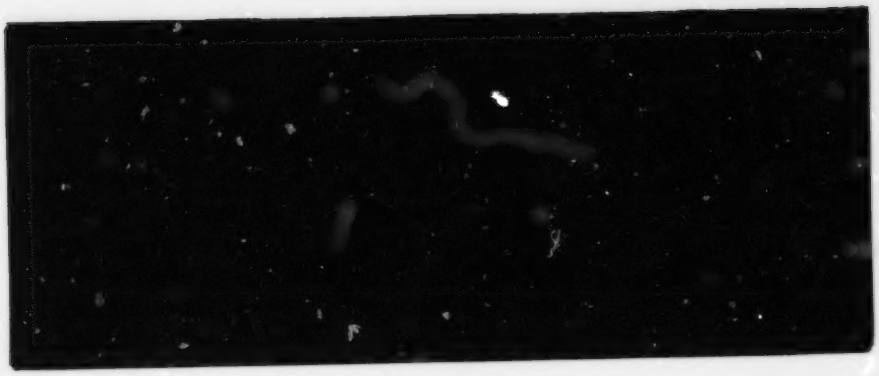

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FINAL REPORT

Dr. P. F. Flood and Dr. S. M. Schmutz

June 1, 1988

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Reproductive Loss in Cattle Caused by Chromosome Abnormalities

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Reproductive Loss in Cattle Caused by Chromosome Abnormalities

a) Abstract

Seventy-five cattle were karyotyped as part of a study of possible causes of subfertility in Saskatchewan beef cattle. Although the 51 bulls represented 8 breeds, most were Charolais. Forty-six bulls had normal chromosomes and 5 had abnormal karyotypes. Three bulls had a 1/29 translocation, one bull had two such translocations, and the fifth bull had a female karyotype.

Twenty-four cows were also karyotyped. Twenty-two because they were relatives of bulls with translocations. Eleven of the cows had normal chromosomes, 9 also had the 1/29 translocation, one had two such translocations, and one cow had three X chromosomes.

It would appear that the incidence of chromosome problems in Saskatchewan beef bulls is relatively high (10%) and if such animals were removed from the breeding population, fertility should improve. The ease with which such a translocation could spread in a herd is demonstrated by the twenty-two female relatives studied, almost half of which are also affected.

Summary

An animal's basic genetic material is carried on structures called chromosomes. Cattle normally have sixty chromosomes. All but the sex chromosomes normally have the centromere (the region of the chromosome which pulls the dividing chromosome into daughter cells) at the end and are therefore referred to as acrocentric chromosomes. Occasionally two chromosomes will fuse (Robertsonian translocation). This results in one chromosome besides the sex chromosomes having a centromere not placed at the end, and also results in a reduction in total number from 60 to 59.

Robertsonian translocations fall into the category of "balanced chromosome rearrangements" because no parts are lost or added, but merely altered in position relative to one another. Balanced rearrangements cause no immediate phenotypic abnormality in the individual which posesses one. Usually, however, reproductive problems ensue (Boue et al. 1985). These lead to subfertility and occasionally infertility. Early embryonic loss often results and can cause prolonged calving intervals, delayed return to estrus and repeapt breeding, low success after artificial insemination, etc. Depending on the chromosomes involved, pregnancies may continue beyond the first three months but give rise to malformed abortuses or calves. This represents an even greater economic loss. Several balanced translocations which cause reduced fertility have been described in cattle (Eldridge 1985, McWhir et al. 1987). Reduced fertility has been demonstrated in carriers of a particularly common translocation of chromosomes 1 and 29 (Gustavsson 1979, Dryendahl and Gustavsson 1979).

Since the risk of this translocation is predicted to be higher in subfertile animals than those of normal fertility and since bulls are used more extensively than individual females, the primary focus of this study was beef bulls rather than cows. The primary objective was to determine if chromosome abnormalities are carried by Saskatchewan beef cattle. Secondarily, we wished to make recommendations to individual owners

regarding the testing of relatives of such translocation carrying animals to assist in eradication. This should ultimately increase the conception rate and decrease embryonic loss in herds with affected animals.

Of the 51 bulls studied, 46 were chromosomally normal and 5 were abnormal (Table 2). Four of the abnormal bulls had a Robertsonian translocation of chromosomes 1 and 29. Two of these four bulls were afterwards found to be brothers and therefore only three different herds were affected. All of the chromosomally abnormal bulls were Charolais. One of the four bulls actually carried two translocation chromosomes which is extremely rare. The fifth animal contains cell lines derived from a female cotwin.

Chromosomes of twenty-four cows were examined as part of this study (Table 3). Twenty-two were Charolais cattle which were relatives of the two translocation carrying bull brothers identified early in the study. Forty-five percent of these cows (10/22) also carried the translocation. In one individual, the translocation appeared twice. Another cow had a karyotype of 61,XXX. Approximately six such individuals have been reported previously (Eldridge 1985).

This study indicates that Saskatchewan beef bulls do unfortunately have chromosome problems. In the subfertile groupd studied here, the incidence of such problems appears to be 10% which is very high. Further study of normal beef bulls will be conducted to determine how widespread this problem is. Since such bulls cause high early embryonic loss, elimination of such translocations should improve herd fertility. The ease with which such a translocation could spread rapidly in a herd is demonstrated by the twenty-two female relatives studied, almost half of which are also affected.

b) technical report -appended

c) Bull's Chromosomes Count (by Connie Lambert)
ADF Newsletter, February 1988
Carolais Banner, April 1988

CBC radio interview of S. M. Schmutz by Rosalie Wolaski
noon farm show, March 1988

Presentation in preparation for Genetic Aspects of Reproduction in Livestock Workshop (satellite meeting of the International Genetics Congress), Aug. 29-30, 1988, Guelph

d) expenditure summary - appended

e) Dr. Peter Flood has spent 5% of his time on this project.
Dr. Sheila Schmutz has spent 70% of her time on this project.
Mrs. Jane Moker has spent 15 hrs./week from Oct.-Mar. on this project and 35 hrs./week in April-May.

f) No item of equipment of greater than $500 in value was purchased.

TECHNICAL REPORT

Dr. S. M. Schmutz and Dr. P. F. Flood

June 1, 1988

Reproductive Loss in Cattle Caused by Chromosome Abnormalities

Abstract

Seventy-five cattle were karyotyped as part of a study of possible causes of subfertility in Saskatchewan beef cattle. Although the 51 bulls represented 8 breeds, most were Charolais. Forty-six bulls had normal chromosomes and 5 had abnormal karyotypes. Three bulls had a 1/29 translocation, one bull had two such translocations, and the fifth bull had a female karyotype.

Twenty-four cows were also karyotyped. Twenty-two because they were relatives of bulls with translocations. Eleven of the cows had normal chromosomes, 9 also had the 1/29 translocation, one had two such translocations, and one cow had three X chromosomes.

It would appear that the incidence of chromosome problems in Saskatchewan beef bulls is relatively high (10%) and if such animals were removed from the breeding population, fertility should improve. The ease with which such a translocation could spread in a herd is demonstrated by the twenty-two female relatives studied, almost half of which are also affected.

Background and Objectives

An animal's basic genetic material is carried on structures called chromosomes. Cattle normally have sixty chromosomes. All but the sex chromosomes normally have the centromere (the region of the chromosome which pulls the dividing chromosome into daughter cells) at the end and are therefore referred to as acrocentric chromosomes. Occasionally two chromosomes will fuse (Robertsonian translocation). This results in one chromosome besides the sex chromosomes having a centromere not placed at the end, and also results in a reduction in total number from 60 to 59.

Such Robertsonian translocations fall into the category of "balanced chromosome rearrangements" because no parts are lost or added, but merely altered in position relative to one another. Balanced rearrangements cause no immediate phenotypic abnormality in the individual which posesses one. Usually, however, reproductive problems ensue (Boue et al. 1985). These lead to subfertility and occasionally infertility. Early embryonic loss often results and can cause prolonged calving intervals, delayed return to estrus and repeapt breeding, low success after artificial insemination, etc. Depending on the chromosomes involved, pregnancies may continue beyond the first three months but give rise to malformed abortuses or calves. This represents an even greater economic loss.

Several balanced translocations which cause reduced fertility have been described in cattle (Eldridge 1985, McWhir et al. 1987). Reduced fertility has been demonstrated in carriers of a particularly common translocation of chromosomes 1 and 29 (Gustavsson 1979, Dryendahl and Gustavsson 1979). Some estimates report that 7% of some purebreed herds may carry this translocation and 8-20% incidence has been reported in the subfertile cohort (Bongso and Basrur 1976, King and

Linnares 1983, Swartz and Vogt 1983, Maurer and Echternkamp 1985). Reduced fertility, particularly among carrier females, has been reported (Refsdahl 1976). No unbalanced liveborn offspring to parents carrying this 1/29 translocation have been reported, but carrier status is transmitted to approximately 50% of the liveborn offspring (Gustavsson 1979).

Since the risk of this translocation is predicted to be higher in subfertile animals than those of normal fertility and since bulls are used more extensively than individual females, the primary focus of this study was beef bulls rather than cows. The primary objective was to determine if chromosome abnormalities are carried by Saskatchewan beef cattle. Secondarily, we wished to make recommendations to individual owners regarding the testing of relatives of such translocation carrying animals to assist in eradication. This should ultimately increase the conception rate and decrease embryonic loss in herds with affected animals.

Experimental Methods

Animals for study were selected because of some evidence of reduced fertility. The criteria used for bulls included a) scrotal circumference less than 32 cm at one year of age, b) low conception rate in cows serviced, or c) semen or sperm abnormality. The criteria for cows included a) known abortions, b) failure to conceive after several months in contact with a bull, or c) animal is a relative of a translocation carrier.

A heparinized blood sample was collected sterilely from each animal. Cultures

were established using 10 ml of Ham's F10 medium (containing 20% fetal calf serum, gentamycin, and fungizone), 8 drops of blood and 0.2 ml of phytohemaglutinin. Cultures were incubated at 37^{0} C in a CO_2 incubator for 3 days. One hour prior to harvest, 0.1 ml of colcemid was added to arrest cell division in metaphase. The cells were exposed to a hypotonic solution (0.75 M KCl) for 45-60 minutes, fixed with a 3:1 solution of methanol: acetic acid overnight and washed 2-3 times before slide preparation. An attempt was made to analyze 15 cells (minimum was 10) from each individual microscopically to rule out mosaicism at the 10% level with 95% confidence (Hook 1977). Photomicrographs were also taken and analyzed.

Results and Discussion

Karyotypes have been prepared and analyzed on 75 cattle. These include 51 bulls and 24 cows. Several chromosomally abnormal animals were identified. In addition 25 other samples were cultured but did not grow. Usually these samples were submitted from veterinarians outside the Saskatoon area. Unfortunately in order to karyotype an animal successfully, a fresh blood sample must be received in the lab and the cells must continue to grow. This necessitates very careful collection and shipment of samples which can be very difficult to achieve from outlying parts of the province.

BULLS

Bulls from eight different breeds were studied (Table 1) although most were Charolais. This bias in Charolais could be caused by several factors. Most animals studied were entered at the R.O.P. test station in Saskatoon and this breed is most common of all breeds represented there (in 1987-1988, 91 of 283 bulls were Charolais). Secondly, since only animals with some evidence of subfertility were sampled, this bias may reflect a slightly lower than average fertility among Charolais bulls compared to others. The former rather than the latter is probably the primary reason for the preponderance of Charolais examined.

Table 1. Breeds of Bulls Karyotyped.

Angus	2
Charolais	31
Hereford	6
Holstein	1
Maine-Anjou	2
Piedmontese	1
Shorthorn	1
Simmental	7

Of the 51 bulls studied, 46 were chromosomally normal and 5 were abnormal (Table 2). Four of the abnormal bulls had a Robertsonian translocation of chromosomes 1 and 29. Two of these four bulls were afterwards found to be brothers and therefore only three different herds were affected. All of the chromosomally abnormal bulls were Charolais.

Table 2. Karyotypes of Bulls.

Normal	60,XY	46
1/29 Translocation	59,XY,t	3

	58,XY,tt	1
Chimera	60,XX/60,XY	1

One of the bulls actually carried two translocated chromosomes which is extremely rare. This phenomenon has only been reported in the Swedish Red and White breed previously (Gustavsson 1969) where the translocation had reached such epidemic proportions that the national government instituted an eradication program.

The fifth animal was presumed to be chimeric. He was born twin to a heifer and all of his blood cells were found to be female rather than male. A skin has been taken from this individual and is under study.

There was no significant difference in scrotal circumference measurements of translocation and normal bulls ($t=-1.85$, $P=0.072$). Therefore given that all the bulls in this study whose circumferences were recorded were under 32 cm, scrotal size is not indicative of cytogenetic abnormality. However, this is not to say that the incidence of translocation is not increased in small testicled bulls since 5/51 (10%) is higher than in the more random study of McWhir et al. (1987) where 2.5 % of 253 cattle had chromosomal abnormalities.

COWS

Twenty-four cows were karyotyped as part of this study (Table 3). Twenty-two were Charolais cattle which were relatives of the two translocation carrying bull brothers identified early in the study. Forty-five percent of these cows (10/22) carried the translocation. In one individual, the translocation appeared twice. Another cow

had a karyotype of 61,XXX. Approximately six such individuals have been reported previously (Eldridge 1985).

Table 3. Karyotypes of Cows.

Normal	60,XX	11
1/29 Translocation	59,XX,t	9
	58,XX,tt	1
Trisomy X	61,XXX	1

The owner of this herd is cooperating fully to eradicate this translocation from his herd. Approximately half of the herd has been tested to date with further testing planned. The fertility is this herd has dropped considerably although not as much as one might predict because the management practice has been to allow the current normal bull a prolonged period of service with the cows (6-8 months) of the year. This has resulted in a long period of calving on this ranch but most cows eventually carried a calf to term. However, the animal with the double translocation was four years of age and had not produced a calf. Only two cows, a Simmental and a Maine-Anjou, were studied because they showed embryonic loss and both were normal.

Conclusions and Impact

This study indicates that unfortunately chromosome abnormalities of the balanced rearrangement type do exist in beef cattle in Saskatchewan. In one purebred Charolais herd the importation of one bull who must have carried the translocation, has caused 50% of the herd to be affected. Several other bulls from the second herd containing a tanslocation bull have been examined and all are normal.

No cows in this herd have been studied to date. The third "herd" has not been studied further because of the recency of detection of this bull. It is unlikely relatives of this bull can be ascertained. Unfortunately he is the double translocation carrier who is 5 years of age and owned by SDA. This means that all calves he has sired will be obligate carriers of the translocation.

The impact of this study is most strongly felt by the Charolais Association since all of the animals with abnormal karyotypes were Charolais. Previous studies in other parts of the world indicate that the continental beef breeds in general are the most likely to be affected (Gustavsson 1979, McWhir et al. 1987). A recent report in the midwestern U.S. also described a relatively high frequency of 1/29 translocations in Charolais (Buoen et al. 1988). The Charolais Association has been contacted regarding the findings of this study and are very interested in collaborating on the eradication of this problem in their breed. Since Charolais bulls are the most sought after bulls to be used in the commercial cross-bred cattle industry in this province it is extremely unfortunate that this translocation seems to occur with rather alarming frequency (4/31=13%) in Charolais bulls with some evidence of subfertility. One can hope that the incidence is much lower in bulls with no evidence of subfertility. This study also emphasizes the importance of continued vigilence regarding bull fertility by private cattle owners and community pastures in the province.

Acknowledgements

We wish to acknowledge the financial support from the Canada-Saskatchewan ERDA agreementand the Canadian Association of Animal Breeders. Expert technical

assistance was given by Jane Moker and Pam Bowman. Blood sampling and reproductive evaluations were kindly performed by Drs. Al Barth, Reuben Mapletoft, Bill Cates, and Eugene Janzen of the Western College of Veterinary Medicine and several private veterianarians.